Comparison of equations to predict the metabolizable energy content as applied to lucerne

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Abstract

Metabolisable energy (ME) intake determines productivity in ruminant production systems and estimates of the ME content of feeds underpin nutritional production models and feeding standards across the world. An inaccurate estimation of ME content of a feed means nutritional models and decision support tools are erroneous, resulting in significant variance in expected liveweight gain or carrying capacity of a pasture. Currently in Australia there are a range of equations used to estimate ME of feeds. Utilising appropriate ME equations suitable for Australian forages, in this case, lucerne, is vital for predictive modelling for production and for any required ration or supplement formulation. The current study compared 24 ME equations in lucerne. Lucerne samples were taken at four different vertical strata grown in field trials of plants of different heights to generate samples of differing nutrient quality.

This research demonstrated that different ME estimation equations generate different ME values for the same lucerne sample. This is exemplified with ME values ranging from 10.25 to 16.58 MJ ME/kg DM for a sample in the top strata, and 7.7 to 13.75 MJ ME/kg DM for another sample in the bottom strata. The Minson (1984) equation, ME (MJ/kg DM) = 0.157 DOMD + 0.059 CP – 1.073, appeared the best equation to use for lucerne according to its lowest SD. This was congruent to the ME equation analysis for forage sorghum samples (Lwin et al. 2022).

This study did not determine which equations were biologically correct, however ME equations based on a combined regression using DOMD and CP parameter were most suitable for use in both forage sorghum and lucerne. This work needs to be validated across multiple forages in Australia to develop ME feeding standards for wider and improved applications for the extensive grazing industry.

Introduction

The ME is a nutritive characteristic and cannot be determined using standard analytical techniques. Feed ME values are estimated via regression equations based on chemical composition (Weiss et al. 1992), estimation of digestibility with *in vitro* methods (Minson 1984; Givens et al. 1990) or gas production methods (Menke &

Steingass 1988; Robinson et al. 2004). These regression equations were originally developed on the basis of calorimetry feeding trials and then related to analytical attributes of a dataset of feeds. Over time, many ME equations have been derived independently using different feed datasets. Equations are then applied, often without consideration of the dataset and parameters with which the equation was determined.

There is limited standardisation of ME equations in Australia. Feeds can be analysed by different methods, using different ME equations through domestic laboratory services but also international laboratory services. Feed ME values can also be obtained by nutritional text feeding tables or using online nutritional tables such as feedipedia. The different equations cause a disparity in ME estimations, particularly in tropical forages. Discrepancy between ME equations was established by Robinson et al. (2004). This work compared and evaluated six ME equations to predict ME based on chemical and *in vitro* components, from US (NRC 2001, University of California at Davis (UC Davis)) and UK (Agricultural Development and Advisory Service - ADAS: Morgan 1972) across a range of feeds. They concluded that no procedure they assessed was able to reliably predict the ME values determined *in vivo* for all feeds. Additional to this in Australia, McLennan (2005) found that ME content estimated by the Australian feeding system standards such as SCA (1990) or Nutrient requirements of domesticated ruminants (NRDR) (Freer et al. 2007) and the Cornell net carbohydrate and protein system (CNCPS) (Fox et al. 2004), which is also commonly used in Australia, differed in tropical forages. Furthermore, Lwin et al. (2022) demonstrated in a study comparing 24 ME equations, that vastly different ME values were generated for each of the 120 forage sorghum samples, and that most equations were not comparable, nor did samples rank similarly across ME equations.

There is a lack of agreement on the appropriate ME equation to use across different environments and production systems in Australia. Livestock production models often use different ME equations to derive intake of ME and predict ruminant production (Robinson et al. 2004). Given the variability between ME equations and lack of information on contextual appropriateness of different models, it is not surprising that these models do not often agree with production results observed in the field. This is particularly so in the predictive productive performance of ruminants fed subtropical forages. Overestimation of ME in a feed will be associated with lower in field production values. This is particularly apparent in tropical forages, as the high NDF content (particularly indigestible NDF) also limits feed intake.

This study aims to compare ME equations to predict the ME content of lucerne. The study objectives were (1) to establish if ME values differed for each lucerne sample and, (2) establish the most appropriate ME predictive equations for lucerne (Best Bet). Appropriate ME assessment will improve the accuracy of ration formulation and production modelling, and integration in existing decision support tools will enable producers to make more informed grazing, supplementation and animal management decisions to maximise productivity.

Methods

Lucerne samples

The lucerne variety, Titan seven was grown at Gatton Research Facility (27°32′45″S, 152°19′44″E) during 2018 and 2019. Different heights of lucerne pasture were sampled. At sampling, plants were harvested 5 cm above the ground, plant height measured, then samples were cut into four equal vertical strata (Benvenutti et al. 2016) tagged and placed into labelled sample bags. Samples were dried in an oven at 60ºC and ground through a 2 mm screen (Retsch Mühle rotary grinder, Germany). A total of 96 samples were selected from a large sample set and used for further analysis. These samples were selected to represent a diverse range of nutritional parameters.

Laboratory analysis

Subsamples were sent to the Dairy One Forage testing laboratory (Ithaca, NY, USA) for nutritional analysis according to CNCPS. Samples were analysed to determine crude protein (CP), ethanol-soluble carbohydrates, lignin, crude fat, acid detergent fiber (ADF), amylase, sodium sulfite treated neutral detergent fiber (NDF) and mineral content by using wet-chemistry services (Dairy One 2007). The Dairy One Forage Lab uses a multiple component summative approach, using total digestible nutrients (TDN) for ME prediction employing a CNCPS approach (Eqn 1 in Table 1).

Subsamples were further analysed locally by using an *in vitro* two-stage rumen fluid pepsin procedure (Tilley and Terry 1963) modified for a Daisy ANKOM system. Estimations were made of dry-matter digestibility (DMD), organic matter digestibility (OMD) and digestible organic matter in the DM (DOMD; Holden 1999). Organic matter (OM) was determined by ashing dried samples at 600°C in a muffle furnace (Modutemp, Midvale, WA, Australia) for 3 hours. Ash-free NDF content was determined according to the method of Goering and Van Soest (1970) modified by Mertens (2002), by using the ANKOM system (ANKOM 200 Fiber Analyzer, Macedon, NY, USA). Other required values for equation application were derived from Dairy One laboratory analysis.

Metabolizable energy equations

A total of 24 equations were used to estimate the ME content in lucerne samples (Table 1). These same equations were applied for the Lwin et al. (2022) study and obtained from a range of Australian, UK and USA feeding standards. All ME equations were utilized for forages.

Table 1. Estimation of ME from different equations in analysis of lucerne samples

Equation number	Author	Equation
Equations based on chemical composition		
1	CNCPS (Fox et al. 2004)	DE (MJ/kg DM) = ((TDN%/100) x 4.409) x 4.184
	NRC (2001)	ME (MJ/kg DM) = ((DE (Mcal/kg DM) x 1.01) – 0.45) x 4.184
2	Minson (1984)	a) ME (MJ/kg DM) = 0.260 CP (%) + 4.653
		b) ME (MJ/kg DM) = 21.574 – 0.207 NDF (%)
		c) ME (MJ/kg DM)) = 16.654 – 0.241 ADF (%)
		d) ME (MJ/kg DM) = 13.764 – 0.165 CP (%) – 0.118 NDF (%)
		e) ME (MJ/kg DM) = 10.738 + 0.161 CP (%) – 0.131 ADF (%)
		f) ME (MJ/kg DM) = 7.735 + 0.17 CP (%) – 0.335 lignin (%)
3	Abate and Mayer (1997)	ME (MJ/kg DM) = 8.11 + 0.1341 CP (%) – 0.1065 ash (%)
Equations based on digestibility		
4	ADAS (Morgan 1972)	ME (MJ/kg DM) = 0.84 + 0.14 DOMD (%)

5	Givens et al. (1990)	ME (MJ/kg DM) = 0.37 + 0.0142 DOMD (g/kg DM) + 0.0077 CP (g/kg DM)
6	Minson (1984)	a) ME (MJ/kg DM) = 0.153 DMD (%) – 1.057 b) ME (MJ/kg DM) = 0.15 OMD (%) – 1.126 c) ME (MJ/kg DM) = 0.184 DOMD (%) – 1.827 d) ME (MJ/kg DM) = 0.157 DOMD (g/100g) + 0.059 CP (%) – 1.073
7	AFRC (Alderman and Cottrill 1993)	ME (MJ/kg DM) = 0.0157 DOMD (g/kg DM)
8	NRDR/CSIRO (Freer et al. 2007)	a) ME (MJ/kg DM) = 0.172 DMD (%) – 1.707 b) ME (MJ/kg DM) = 0.169 OMD (%) – 1.986 c) ME (MJ/kg DM) = 0.194 DOMD (%) – 2.577
9	AFIA (2011)	ME (MJ/kg DM) = 0.203 DOMD (%) – 3.001
10	SCA (1990)	a) ME (MJ/kg DM) = 0.18 DOMD (%)– 1.8 b) ME (MJ/kg DM) = 0.16 OMD (%)– 1.8 c) ME (MJ/kg DM) = 0.17 DMD (%) – 2.0
11	Freer et al. (2004)	ME (MJ/kg DM) = 0.172 DMD (%) – 1.71

Where; Dry matter digestibility (DMD) = (feed DM – residual DM)/feed DM

Organic matter digestibility (OMD) = (feed OM – residual OM)/feed OM

Digestible organic matter in DM (DOMD) = (feed OM – residual OM)/feed DM

Statistical analyses

The set of predicted ME values generated by the 24 equations for each of the 96 lucerne samples underwent a series of analyses. Firstly, an ME index was calculated to account for each height stratum within each sample. The ME index was calculated by ranking from lowest to highest, the average ME values for each stratum within each sample across all ME equations. The predictions generated by the different ME equations were then regressed against the ME index by fitting linear mixed effects (LME) models. Model comparisons using Akaike, and Bayesian information criterions indicated that a random slope and intercept structure was optimal. The set of ME equations was narrowed by selecting only those equations whose random slope and intercept fell within the 95% confidence interval of the overall LME fixed effect slope and intercept, i.e., those equations that generated ME predictions closest to the overall mean predictions across all ME equations combined. The 'optimal' ME equation was then identified by selecting the equation from the narrowed subset of equations with the lowest standard deviation of its predictions, i.e., had the lowest variability across the range of ME values tested.

Results

The 24 ME equations had variable ME predictions for each lucerne sample. The full set of 24 ME equations was narrowed to eight preferred equations based on their random slopes and intercepts falling within the 95% confidence interval of the overall fixed effect slope and intercept. These eight equations were all based on digestibility, whether as a sole parameter or digestibility combined with a CP parameter. (equation 6d, 8a, 8b, 8c, 9, 10b, 10c and 11). When SD were taken into account equations: 10b (SCA 1990) was considered Best Bet for the top strata, 6c (Minson 1984) for strata 2, 6b (Minson 1984) for strata 3 and equation 5 (Givens et al. 1990) for the bottom strata. This is not practical to have different equations preferred for the various strata and as such a preferred equation was considered across all strata. These equations that were within the 95% Cl of both fixed effect slope and intercept were then ranked using standard deviations (SD) (Figure 1). The equation with the lowest SD was selected as the Best Bet ME equation for lucerne. The ME equation with the lowest SD was Eqn 6d (ME (MJ/kg DM) = 0.157 DOMD (%) + 0.059 CP (%) – 1.073) (Minson 1984).

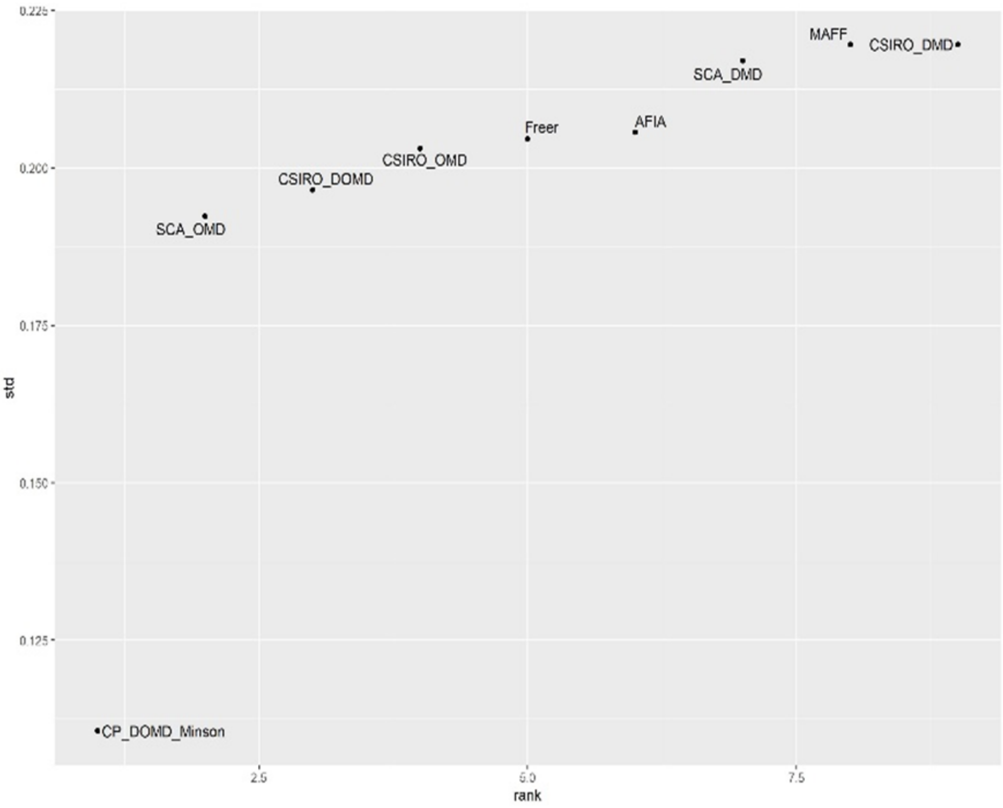

Figure 1. Equations ranked by standard deviations (SD) from lucerne sample dataset. Equations are: CP_DOMD_Minson = Equation 6d, SCA_OMD = Equation 10b, CSIRO_DOMD = Equation 8c, CSIRO_OMD = Equation 8b, Freer = Equation 11, AFIA = Equation 9, SCA_DMD = Equation 10c, MAFF = Equation 9, CSIRO_DMD = Equation 8a. Each equation (Eqn) is defined in Table 1.

Discussion

This research focused on the estimation of ME in lucerne as a model crop for legumes. The ME value is the first limiting nutrient and determines the maximum productivity of the ruminant. The content of ME in feed is most accurately measured via ruminant calorimetry studies (Blaxter & Clapperton 1965) but this is not practical for a feed analysis measurement. As such ME equations have been derived from regression relationships between digestibility or chemical composition with ME values. It is assumed that this relationship varies between feeds and that ME can be predicted satisfactorily by different laboratory methods (Minson 1980). An accurate estimation of ME content is fundamental to accurate prediction of productive performance of ruminants and to better assist producers to make grazing management decisions. The current study compared 24 equations to estimate the ME content in a lucerne dataset of 96 samples. Lucerne samples were taken at four different vertical strata grown in field trials containing plants of different heights. The ME equations were that used by Lwin et al. (2022) and was not an exhaustive list but rather a selection of equations that are utilized in recognized feeding systems. These equations selected were either developed using chemical composition, digestibility data, or a combination of both chemical composition and digestibility data. It was also important to have knowledge regarding the equations associated feed databases such as the number of feeds and type of feeds. Only equations derived from forage databases were utilized.

Our research shows for each individual lucerne sample, different ME estimation equations will give vastly different ME values. Similarly, Lwin et al. (2022), using the same equations for comparison, observed very different ME estimations on individual forage sorghum samples. In many of these samples, ME estimations were not biologically sensible (over 17 MJ/kg DM). This variability in estimations was even greater in higher quality lucerne (top strata 1 and 2) compared to lower quality forage (bottom strata 3 and 4). This variability between ME estimations for the same sample in this research using legumes and also in Lwin et al. (2022) using a topical grass, exemplifies the importance of utilizing appropriate ME equations. Utilising an ME equation that is not suitable for a feed type would likely provide incorrect ME values. It is also imperative to know the derivation of feed ME values (when using feed laboratories or feed table values) when comparing different feeds e.g. for ration formulation, as this research has shown that there will be major inconsistencies when comparing ME values from different equations.

As with the Lwin et al. (2022) equation analysis for forage sorghum, this study with lucerne samples, could not definitively determine which equation was biologically correct, however through a series of statistical approaches, these equations were compared and Best Bet equations were ascertained. For lucerne, ME equations using a digestibility parameter provide acceptable ME estimations compared to those equations based solely on chemical composition. In particular, the equations based on a combined regression using the parameters digestible organic matter in the DOMD and CP were most suitable for use, The predicted ME equation from Minson (1984) based on CP and DOMD is the best equation to use for lucerne according to its lowest SD which is ME (MJ/kg DM) = 0.157 DOMD + 0.059 CP – 1.073. These results are congruent to the ME equation analysis for the forage sorghum study.

Utilizing digestibility as a parameter in an ME estimation equation is biologically appropriate. ME is the energy in the feed remaining after subtracting the energy of the faeces, urine and combustible gases such as methane. There is a biological correlation of ME with digestibility (Alderman and Cottrill 1993; Freer et al. 2007). Minson (1984) further discussed that ME has a high correlation with DMD and OMD with lower error compared to feed ME estimations from chemical composition when working with *Digitaria setivalva*. Similarly, Armstrong (1964) noted less SD of ME estimation from digestibility based values compared to utilizing chemical composition attributes in sixteen grasses.

The inclusion of CP as a parameter is also significant. The calorific value of digestibility of OMD has a significant relationship with CP as higher digestibility occurs with increasing proportion of CP in forages (Terry et al. 1974) attributable to a higher N supply for microbial activity (Satter and Slyter 1974). The protein also when broken down is a further source of energy to the animal that needs to be accounted for. Lwin et al. (2022), in an analysis of 24 ME equations found, using forage sorghum as a sample set, the Best Bet equations were DOMD and CP from Givens et al. (1990) and Minson (1984). As such, based on the current research, ME equations which utilized the parameters of DOMD and CP can be used universally in both tropical grasses and lucerne. Moreover, the Best

Bet equation from CP and DOMD in Minson (1984) can easily be analyzed in the laboratory and also obtained through faecal NIRS estimates from rangeland animals.

Conclusion and implications

There is a need for agreement on the appropriate ME equation to use in various production systems, as this will improve the accuracy of ration formulation, and importantly for grazing management decisions and livestock production modelling.

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